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Seroprevalence of CMV in Women with Bad Obstetric History in Babil/Iraq

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Abstract

Placental dysfunction and fetal central nervous system infestation caused by Human cytomegalovirus (HCMV) is the leading cause of congenital non-genetic neuro-developmental problems of the newborn worldwide. Although the highest rates of congenital infection and CMV seroprevalence occurs in developing countries like Iraq, there remains a paucity of data from that part of the world. This descriptive case control study was undertaken in Babylon/ Iraq to determine the local seroprevalence of CMV in women of child bearing age and to identify the socio-demographic factors associated with it. This study found a seropositivity peak amongst the 26-35 yr olds which declined in the 36 – 45 yr olds. However, the evidence of current infection was stable at 25% among the 26-35 yr olds and the 36 – 45 yr old women. Overall, seropositivity was at 77.32%, a susceptibility rate was at 22.68%, and seropositivity for IgG was highest among the educated, those living in overcrowded settings, and those with poor obstetric histories. Our study concludes that CMV screening of women in the Al Hamza district in Babylon/Iraq and the availability of advice on how to prevent the infection can be beneficial for health outcomes.

Keywords: CMV, Cytomegalovirus, women, ELIZA, poor obstetric history, Iraq, Babil

الانتشار المصلي للفيروس المضخم للخلايا لدى النساء ذوات تاريخ الولادة السيئ في بابل / العراق

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الخلاصة

يعد الخلل الوظيفي في المشيمة و / أو إصابة الجهاز العصبي المركزي للجنين بسبب الفيروس المضخم للخلايا البشري (HCMV) السبب الرئيسي لمشاكل النمو العصبية الخلقية غير الوراثية للمواليد في جميع أنحاء العالم على الرغم من أن أعلى معدلات العدوى الخلقية والانتشار المصلي للفيروس المضخم للخلايا تحدث في البلدان النامية مثل العراق ، لا يزال هناك ندرة في البيانات من هذا الجزء من العالم. أجريت هذه الدراسة الوصفية للشواهد في بابل / العراق لتحديد الانتشار المصلي المحلي لفيروس CMV في النساء في سن الإنجاب ، وللتعرف على العوامل الاجتماعية والديموغرافية المرتبطة به. وجدت هذه الدراسة ذروة الإيجابية المصلية بين الأشخاص الذين تتراوح أعمارهم بين ٢٦ و ٣٥ عامًا والتي انخفضت في الفئة العمرية ٣٦-٤٥ عامًا. ومع ذلك ، فإن الأدلة على الإصابة الحالية كانت مستقرة عند ٢٥ ٪ بين ٢٦-٣٥ سنة والنساء بين ٣٦ - ٤٥ سنة. كانت الإيجابية المصلية الإجمالية عند ٧٧,٣٢٪ ، ومعدل الحساسية ٢٢,٦٨٪ ، والإيجابية المصلية لـ IgG كانت الأعلى بين المتعلمين ، والذين يعيشون في ظروف مزدحمة ، وأولئك الذين لديهم تاريخ توليدي ضعيف. خلصت دراستنا إلى أن فحص الفيروس المضخم للخلايا للنساء في منطقة الحمزة في بابل / العراق وتوافر النصائح حول كيفية الوقاية من العدوى يمكن أن يكون مفيدًا للنتائج الصحية.

الكلمات المفتاحية: الفيروس المضخم للخلايا ، النساء ، اليزا ، تاريخ الولادة الضعيف ، العراق ، بابل

Introduction

Worldwide, 0.1-0.2% of all pregnancies result in permanent disabilities of varying degree because of active HCMV infection. Disabilities caused by HCMV in the newborn include blindness, deafness, microcephaly, cerebral palsy, intellectual disability, developmental delay, and in rare cases death [(1).] Such disabilities may be lifelong and / or severe. HCMV is the leading cause of infectious (congenital) neurologic handicap in the newborn [(2)]. Globally, 0.5 – 1.1% of all pregnancies occur in women with active HCMV infection. Some of these infections are a reawakening of dormant historic episodes. 90% of these pregnancies result in the birth of asymptomatic babies, however, 5-10% of babies who are asymptomatic at birth eventually die of HCMV related defects [(3)].

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Epidemiology

Globally, there is epidemiologic variation in HCMV infection in women of childbearing age; varying from 30.4% (in Ireland) to 98.9% (in Turkey) (4, 5). The seroprevalence rate in women with a bad obstetric history (BOH) varies from 14.2% (Iran) to 91.05% (India) (6, 7). Amongst Arab countries, seroprevalence in pregnant women varies from 77.8% (Iraq) to 88% (Jordan) (8, 9). For Arab women with BOH, it varies from 4.8% (Iraq) to 95% (Jordan) (10).

Study aims and objectives

Although there is an accumulation of information on HCMV seroprevalence worldwide, there is however little data informing the seroprevalence of HCMV local levels in Iraq. The objective of the current study is to investigate the seroprevalence of CMV in women of child bearing age with a BOH in the Al Hamza district in Babylon/Iraq and the socio-demographic variables which might affect seropositivity.

Materials and Methods

Study design

This is a descriptive case—control study. The total study population involves *(n=115)* women of childbearing age (15-45 years old). To ensure a good spread of the subjects, women with bad obstetric history were identified and recruited from the private labs and the Primary Healthcare Centers situated in urban and rural areas in the Al Hamza district in Babylon Governorates. Twent-five of the women were excluded as they did not meet the study criteria and had missing data. Of the remaining *(n=90)* members of the study population, 54 (60%) had BOH and considered as study group subjects while the 36 (40%) had average to good obstetric history and considered as control group subjects.

Collection of data

Members of the project team visited primary health care centers and the private labs in the region on a regular basis. Possible candidates were identified, approached, interviewed and were suitable, included in the study population once informed consent and permission had been sought and obtained. Four groups of patients were recruited.
Group 1 – Pregnant women with average to good obstetric history.
Group 2- Pregnant women with bad obstetric history.
Group 3 – Non-pregnant women with average to good obstetric history.
Group 4- Non-pregnant women with bad obstetric history.
Subjects were interviewed to exclude those whose BOH was due to other causes e.g. uterine or cervical abnormalities (e.g. cervical incompetence), chromosomal abnormalities in either spouse, poorly controlled hyperthyroidism, diabetes mellitus, hypertension, hypothyroidism, renal disorders, a history of foetal chromosomal or genetic anomalies.

Collection of blood samples:

Blood samples were collected from January 2018 until December 2019. Around 5—10 mL of blood was collected from vein for each case and placed in a sterile container with strict aseptic conditions. The serum was then separated and stored in aliquots at −20 ◦C until further processing.

ELISA Essay

Serum samples collected from the study and control groups subjects were diagnosed for CMV IgM and IgG antibodies using Cyto Megalo Virus ELISA kits which are commercially available. The kit was purchased from CALBIOTECH, CM028M and CM028G for IgM and IgG respectively. ELISA test was performed according to the manufacturer's instructions (CALBIOTECH). The results were processed using a microwell ELISA reader and read at an optical density 450 nm.

Data analysis

Using Microsoft excel spreadsheet, the collected data were compiled, assembled and the odds ratios computed for suitable analysis. Using chi squared test via SPSS (V.24), the association between categorical data was examined. Accuracy, especially in instances of smaller sample sizes, was ensured using power analysis. Association between variables was determined using bivariate regression-line analysis to compute odds ratio. CMV infection determinants were ascertained through regression line analysis and odds ratio, while standardising for confounding factors such as age, residential or educational status.

Results

A total of *(n=115)* women were identified and pooled for this study. Incomplete data were recorded for 25 of the women in the study sample. The women in the sample were categorized into further subgroups using different factors such as education level, pregnant or not pregnant, residential status (overcrowding or not overcrowded), bad obstetric history or normal obstetric history and whether or not they aborted (Table 1).

Table 1. Sample population with recorded versus missing IgG and IgM results

	Data Summary					
	Valid		Missing		TOTAL	
	N	**Percent**	**N**	**Percent**	**N**	Percent
Sample population with recorded v missing CMV_IgM result.	90	78.3%	25	21.7%	115	100%
Sample population with recorded v missing CMV_IgG result.	90	78.3%	25	21.7%	115	100%

Of the 90 women whose data was complete, 34 were in the 15-25 yr old age group, 48 in the 26-35 yr old age group, and 8 in the 36 – 45 yr age group. 23 of the 34 (67%) of the 15-25 yr age group were positive for IgG, 40 of the 48 (83%) of the 26-35 yr age group were positive for IgG, and 6 of the 8 (75%) of the 36-45 yr age group were positive for IgG (Table 2).

Table 2. Prevalence of CMV IgG among different age groups

Age range	**CMV_IgG**		Total
	Negative	**Positive**	
15-25 yr old women	11.00	23.00	34.00
	32.35%	67.65%	100.00%
26-35 yr old Women	8.00	40.00	48.00
	16.67%	83.33%	100.00%
36-45 yr old Women	2.00	6.00	8.00
	25.00%	75.00%	100.00%
Total	21.00	69.00	90.00
	23.33%	76.67%	100.00%

For IgM, 7 of the 34 (20.59%) of the 15-25 yr age group were positive for IgM, 12 of the 48 (25%) of the 26-35 yr age group were positive for IgM, and 2 of the 8 (25%) of the 36-45 yr age group were positive for IgM (Table 3).

Table 3. Prevalence of CMV IgM among different age group

Age range	***CMV_IgM***		Total
	Negative	**Positive**	
15-25 yr old women	27.00	7.00	34.00
	79.41%	20.59%	100.00%
26-35 yr old Women	36.00	12.00	48.00
	75.00%	25.00%	100.00%
36-45 yr old Women	6.00	2.00	8.00
	75.00%	25.00%	100.00%
Total	69.00	21.00	90.00
	76.67%	23.33%	100.00%

There were age disparities in the distribution of IgG and IgM seroprevalence among the study population. Starting at 33.33% in the 15-25 yr old, IgG seroprevalence peaked at 57.97% in the 26-35 yr olds before declining to 8.7% in the 36-45 yr old. A similar pattern is seen for IgM starting at 33.33% amongst the 15-25 yr old, peaks at 57.14% in the 26-35 yr old and declining to 9.52% in the 36-45 yr old.

Table 4. Seroprevalence of CMV IgG and IgM in regards to pregnancy, abortion, education, BOH and residential status

	Age Groups			BOH		Pregnancy status		Residential status		Education status		Aborted	
	15-25 yr. old women	26-35 yr. old women	36-45 yr. old women	Normal pregnancy	BOH	Not Pregnant	Pregnant	no over-crowding	Over-crowding	Un-Educated	Educated	Not Aborted	Aborted
CMV_IgM NEGATIVE	27.00	36.00	6.00	34.00	39.00	27.00	44.00	8.00	63.00	11.00	52.00	35.00	39.00
	39.13%	52.17%	8.70%	46.58%	53.42%	38.03%	61.97%	11.27%	88.73%	17.46%	82.54%	47.3%	52.7%
CMV_IgM POSITIVE	7.00	12.00	2.00	5.00	18.00	13.00	9.00	1.00	20.00	2.00	19.00	5.00	18.00
	33.33%	57.14%	9.52%	21.74%	78.26%	59.09%	40.91%	4.76%	95.24%	9.52%	90.48%	21.74%	78.26%
Pearson x2 value	0.23			**4.47**		3.04		0.78		0.76		0.65	
P value	>0.05			**0.034**		>0.05		>0.05		>0.05		>0.05	
CMV_IgG NEGATIVE	11.00	8.00	2.00	15.00	6.00	11.00	10.00	2.00	20.00	4.00	13.00	16.00	6.00
	52.38%	38.10%	9.52%	71.43%	28.57%	52.38%	47.62%	9.09%	90.91%	23.53%	76.47%	72.73%	27.27%
CMV_IgG POSITIVE	23.00	40.00	6.00	24.00	51.00	29.00	43.00	7.00	63.00	9.00	58.00	24.00	51.00
	33.33%	57.97%	8.70%	32.00%	68.00%	40.28%	59.72%	10.00%	90.00%	13.43%	86.57%	32.00%	68.00%
Pearson x2 value	2.75			**10.57**		0.97		0.02		1.06		**11.64**	
P value	>0.05			**0.001**		>0.05		>0.05		>0.05		**0.001**	
Bold values indicated a significant difference													

Overall, the seroprevalence of CMV within the study sample was 77.32%. IgG levels were highest among those living in overcrowded situations (90%), those who had abortions (68%), and the educated (86.57%). Interestingly, IgG levels were highest (68%) among those with a poor obstetric history (study group), compared to 32% for those with moderate to good obstetric history (control group). Evidence of current infection, i.e. positive IgM levels was highest amongst those in overcrowded situations (95.24%), those with a poor obstetric history (78.26%), those who had abortions (78.26%), and the educated (90.48%).

Contrary to other studies the seroprevalence of IgM amongst pregnant women (40.91%) was lower than in the women who were not pregnant (59.09%). The converse was true of IgG levels being 59.72% (pregnant) and 40.28% (not pregnant) (Table 4).

The chi squared values and p-values indicate that x^2 figures for women with a bad obstetric history, both for IgG and IgM was significant at a p value of below 0.05 at 95% confidence interval (CI). This was also true of those women with aborted fetus. This association was also confirmed by the odds ratio, the x^2 BOH * CMV_IgM was 4.47 *(p value = 0.034)* while the x^2 of BOH * CMV_IgG was 10.57 *(P value=0.001)* and the x^2 of CMV_IgG * Abortion was 11.64 with *(P value=0.001*). There is however no significant correlation *(P value>0.05)* found between CMV_IgG/ IgM and Residency, Educational level, age group and pregnancy status.

Discussion

According to the WHO [11], the seroprevalence of HCMV in women of child bearing age in the European region is 63-76%, this is lower than that in the East Mediterranean region (88-95%), the region of the Americas (69-87%), the west Pacific region (86-94%), the African region (82-94%), and the South east Asian regions (82-94%) [12]. Other studies undertaken in other parts of Iraq have found seroprevalence rates in the region of 95.7% in Kirkuk [13]. The overall seroprevalence of CMV within this study sample was 77.32%, this study found HCMV seroprevalence highest amongst 26-35 yr old women but declining with age. This study found a susceptibility rate of 22.68%, which is higher than the average for the country. The study found an association of current infection with abortion and BOH (Control group). Other findings of this study include the higher rates of seroprevalence amongst the educated and among those living in overcrowded households.

The higher seroprevalence amongst 26-35 yr old is aligned with previous studies which have attributed this to the exposure of these women to school age children, especially as their own children begin to attend school [14]. The finding of higher rates amongst the educated and amongst those living in overcrowded households is also in alignment with previous studies [15]. The overall prevalence of 77.32% and the susceptibility rate of 22.68% highlights the need for routine screening in antenatal clinics. This study shows that while the average rates of CMV seroprevalence in Iraq may be over 95% [12], there are regional variations which require local adjustment of national policies on CMV infection prevention and management. Scholars have argued against routine screening because of high levels of seroprevalence, cost, extensive viral strain diversification, and the inconsistent and ineffective treatment options available for HCMV [16]. However, the arguments against routine screening, awareness promotions, prevention campaigns, and treatment may be predicated on a general underestimation or a lack of understanding of the enormity of the challenge posed by HCMV infection as a disease burden.

HCMV is highly adapted to its human host, it is a member of the HSV group. Primary infection with the virus results in an initial period of viral replication and shedding in body fluids including saliva, breast milk, urine, and in genital secretions. This is followed by a viraemia and, in some people, an infectious mononucleosis phase. Finally, the host develops a generalized immune response involving the entire immune system. Thereafter the infection enters a latent phase when viral levels are either low or even absent to detection but present in $CD14^+$ and also in the $CD33^+$ & 34^+ cells of the mononuclear cells of the peripheral blood and bone marrow respectively. These sites then serve as reservoirs for future reactivation at opportune moments [17]. This cycle perpetuated by the virus accounts for the delayed onset of symptoms in congenitally infected babies, the initial asymptomatic infection in many, and therefore, the presumption in many quarters that babies born to mothers with prior HCMV antibodies have normal outcomes. Yet nothing could be further from the truth. The diversification of the virus creates a potential new threat to those who are already seropositive. The fact that the source of such diversity has been found to be endogenous makes those who are seropositive or living in a high seroprevalence environment particularly at risk [18]. It is not surprising therefore, that the greatest burden of HCMV burden is found in developing countries where there is also a high level of seropositivity. High viral diversification continues to present challenges to the development of effective vaccines. A challenge which is amplified by the fact that large proportions of congenitally infected babies are the product of mothers with demonstrable HCMV antibodies [19].

The consistent finding of an association between poorer socioeconomic conditions such as overcrowding, breast milk transmission and high HCMV seroprevalence offers an opportunity to

address this problem while an effective vaccine is awaited [15]. Contrary to previous conception that higher seroprevalence confers immunity, the high viral diversity of HCMV predisposes the already seronegative women to reinfection with a different strain from the community or from endogenous viral reactivation. Thus, higher seroprevalence may actually predispose to higher likelihood of infection. Therefore, the higher seroprevalence of HCMV in the rural and/or urban areas of Babylon should not occasion complacency to prevention and treatment but rather is the more reason for intervention.

Educating women and children about HCMV preventive measures is known to be effective [20]. Basic preventive measures like: Teaching people to wash their hands regularly, catch their sneezes and body fluids and dispose of this in the appropriate way, avoiding the sharing of cutleries, plates and cups, educating mothers not to put babies' pacifiers in their mouths if it has been in a child's mouth, or advising mothers to avoid kissing children on the lips, have been shown to be feasible practices observable by mothers when adopted and effective in limiting the spread of infection. Other measures include the introduction and reinforcement of these messages in antenatal clinics, children's clinics, and hospitals using relevant staff holding classes and demonstrations with women and their husbands. There is also an opportunity to use billboards or posters in the hospital to adorn the walls of the waiting rooms or consulting rooms and wards where women are likely to see them. All such posters, classes, advice, and discussions should take place and employ cultural and language sensitive media, observing the complexities and subtleties of local customs and practices to drive home the message.

Conclusion

This is the first study into the seroprevalence of HCMV amongst women of childbearing age in Al-Hamza district in Babylon/Iraq. The study found that a significant number of women were susceptible to HCMV infection, this finding contrasts the average of over 95% seroprevalence rate for the country. This study highlights the need, as the search for an effective vaccine and the debate over the cost benefit of routine screening continues, for seroprevalence data of HCMV in local populations to be researched and documented. This is because there is a need for age stratified, population based HCMV seroprevalence data in the planning of demographically appropriate modelling and strategic intervention programs for local HCMV infection in the Al Hamza district in Babylon/ Iraq. Bridging such a gap is the reason for this paper.

References

1. NSW. Cytomegalovirus (CMV) and pregnancy fact sheet. Available at: https://www.health.nsw.gov.au/Infectious/factsheets/Pages/cmv-and pregnancy.aspx#:~:text=CMV%20is%20a%20common%20viral,while%20others%20are%20born%20healthy; 2017 {accessed 22.06.2020}.
2. Walker, S.P., Palma-Dias, R., Wood, E.M. et al. Cytomegalovirus in pregnancy: to screen or not to screen. BMC Pregnancy Childbirth. 2013; 13, 96. https://doi.org/10.1186/1471-2393-13-96. {Accessed 23.06.2020}.
3. Dollard, S.C., Grosse, S.D., Ross, D.S. New estimates of the prevalence of neurological and sensory sequelae and mortality associated with congenital cytomegalovirus infection. Rev Med Virol. 2007;17(5):355–363.
4. Knowles, S.J., Grundy, K.C., Cafferkey, M.T., Cahill, I., Geary, M.L. Low cytomegalovirus seroprevalence in Irish pregnant women. Ir Med J. 2005;98:210—2.
5. Koksaldi-Motor, V., Evirgen, O., Azaroglu, I., Inci, M., Ozer, B., Arica, S. Prevalence of toxoplasmosis, cytomegalovirus and rubella IgG antibodies in Hatay women and children. West Indian Med J. 2012;61(2):154—7.
6. Falahi, S., Ravanshad, M., Koohi, A.K., Karimi, A.M. Short com-munication: seroprevalence of CMV in women's with spontaneous abortion in kowsar hospital, Ilam during 2007—2008. Modares J Med Sci Pathobiol. 2010;12:39—43.
7. Turbadkar, D., Mathur, M., Rele, M. Seroprevalence of TORCH infection in bad obstetric history. Indian J Med Microbiol. 2003;21:108—11, 2003.
8. Daboubi, M. and Al-Zaben, S. Cytomegalovirus infection inwomen of childbearing age in Jordan. Jordan Med J. 2000; 34:106—8.
9. Abdul Mohymen, N., Hussien, A., Hassan, F.K. Association between TORCH agents and recurrent spontaneous abortion. Iraqi J Med Sci. 2009; 7:40—6.
10. Al-Marzoqi, A.H.M., Kadhim, R.A., Aljanabi, D.K.F., Hussein, H.J., AlTae, Z.M. Seroprevalence study of IgG and IgM antibodiesto toxoplasma, rubella, cytomegalovirus, Chlamydia tra-chomatis and Herpes simplex II in Pregnancy women inBabylon Province. J Biol Agric Healthc. 2012;2:159—64.
11. Nations U. World Population Prospects. revision. https://www.un.org/development/desa/publications/world - population - prospects - the - 2017 - revision.html; 2017 {Accessed on 18.01.20}.
12. Zuhair, M., Smit, G.S..A, Wallis, G., et al. Estimation of the worldwide seroprevalence of cytomegalovirus: A systematic review and meta - analysis. Rev Med Virol., Available at:

https://doi.org/10.1002/rmv. 2019; 29(3):e2034 {Accessed on 12.10.20}.

13. Aljumaili, Z.K.M., Alsamarai, A.M., Najem, W.S. Cytomegalovirus seroprevalence in women with bad obstetric history in Kirkuk, Iraq. Journal of Infection and Public Health. 2014; 7, 277—288.

14. Cannon, M.J., Schmid, D.S., Hyde, T.B. Review of cytomegalovirus seroprevalence and demographic characteristics associated with infection. Rev Med Virol. 2010;20(4):202 - 213., 20(4):202 - 213.

15. Pembrey, L., Raynor, P., Griffiths, P., Chaytor, S., Wright, J., Hall, A.J. Seroprevalence of cytomegalovirus, Epstein Barr virus and varicella zoster virus among pregnant women in Bradford: a cohort study. PLoS One.2013; 8(11):e81881.

16. Manicklal, S., Emery, V. C., Lazzarotto, T., Boppana, S. B., & Gupta, R. K. The "silent" global burden of congenital cytomegalovirus. Clinical microbiology reviews. 2013; 26(1), 86–102. https://doi.org/10.1128/CMR.00062-12

17. Mocarski, J.E., Shenk, T., Pass, R. Cytomegaloviruses, p 2702–2772. In Knipe D, Howley P (ed), Fields virology, 5th ed. Lippincott Williams and Wilkins, Philadelphia, PA. 2007; p 2702–2772.

18. Arora, N., Novak, Z., Fowler, K.B., Boppana, S.B., Ross, S.A. Cytomegalovirus viruria and DNAemia in healthy seropositive women. J. Infect. Dis. 2010; 202:1800 –1803.

19. Renzette, N., Bhattacharjee, B., Jensen, J.D., Gibson, L., Kowalik, T.F., Extensive genome-wide variability of human cytomegalovirus in congenitally infected infants. PLoS Pathog. 2011; 7:e1001344. doi:10.1371/journal .ppat.1001344.

20. Adler, S.P., and Nigro, G. Prevention of Maternal-Fetal Transmission of Cytomegalovirus. CMV Transmission, CID. 2013; 2013:57 (Suppl 4) • S189.

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